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Relation of Water Loss to the Hatching of Eggs from Detached Oöthecae of *Blattella germanica* (L.)[1]

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The females of the German cockroach, *Blattella germanica* (L.), carry their oöthecae externally until or shortly before the eggs hatch. According to Ross (1929) when 9-day old or older oöthecae were removed from females, the eggs could hatch if the egg cases were kept under cool humid conditions; however, when oöthecae younger than 9 days were removed, the eggs failed to hatch under any conditions. Woodruff (1938) noticed that a green band appeared in the alimentary tract (which can be seen through the wall of the oötheca) of embryos of *B. germanica*, usually 3 days before hatching; he claimed that the eggs in oöthecae removed from females prior to the appearance of this band failed to hatch. Parker & Campbell (1940) found that the green band may first be observed 6 to 7 days before the eggs hatch (17 to 28 days at 76° to 83° F.). However, they found that some eggs hatched from oöthecae which were removed from females long before the appearance of the green band in the embryos. Some of the eggs in one oötheca hatched 24 days after being detached from the female which would indicate that this oötheca was fairly young at the time of its removal. In this paper we present some data which may explain the above discrepancies in hatching of eggs from detached oöthecae.

The cockroaches were reared on Purina dog chow checkers. Females that were forming oöthecae were isolated every 24 hours and kept, until used, at 27° to 29° C. with food and water. The insects were anesthetized with CO_2 when the oöthecae were manually removed. The egg cases were weighed on a torsion balance that was sensitive to 0.05 mg. Oöthecae were air-oven dried at about 80° C. to constant weight.

In table 1 and figure 1A and 1B is shown the effect of relative humidity on the hatching of eggs from detached oöthecae of *B. germanica*. The eggs in oöthecae kept at a low humidity of 30 per cent to 50 per cent did not hatch unless they had almost completed their development before being detached; at the low humidity the percent-

Table 1.—Effect of relative humidity on hatching of eggs from manually detached oöthecae of *Blattella germanica*.

Age (Days) of Oöthecae When Detached	Oöthecae Kept at 27°–29° C. and:			
	30%–50% R. H.		90% R. H.	
	Number Used	Number from Which Some Eggs Hatched	Number Used	Number from Which Some Eggs Hatched
1	20	1	34	16
2	20	0	29	1
3	22	0	31	0
4	21	0	47	1
5	20	0	32	1
6	27	0	23	1
7	40	0	28	10
8	35	1	32	22
9	17	0	20	17
10	20	2	27	24
11	29	3	17	16
12	69	3	33	26
13	40	4	22	20
14	50	9	22	22
15	61	9	10	10
16	34	31		
Total	525	63	407	187

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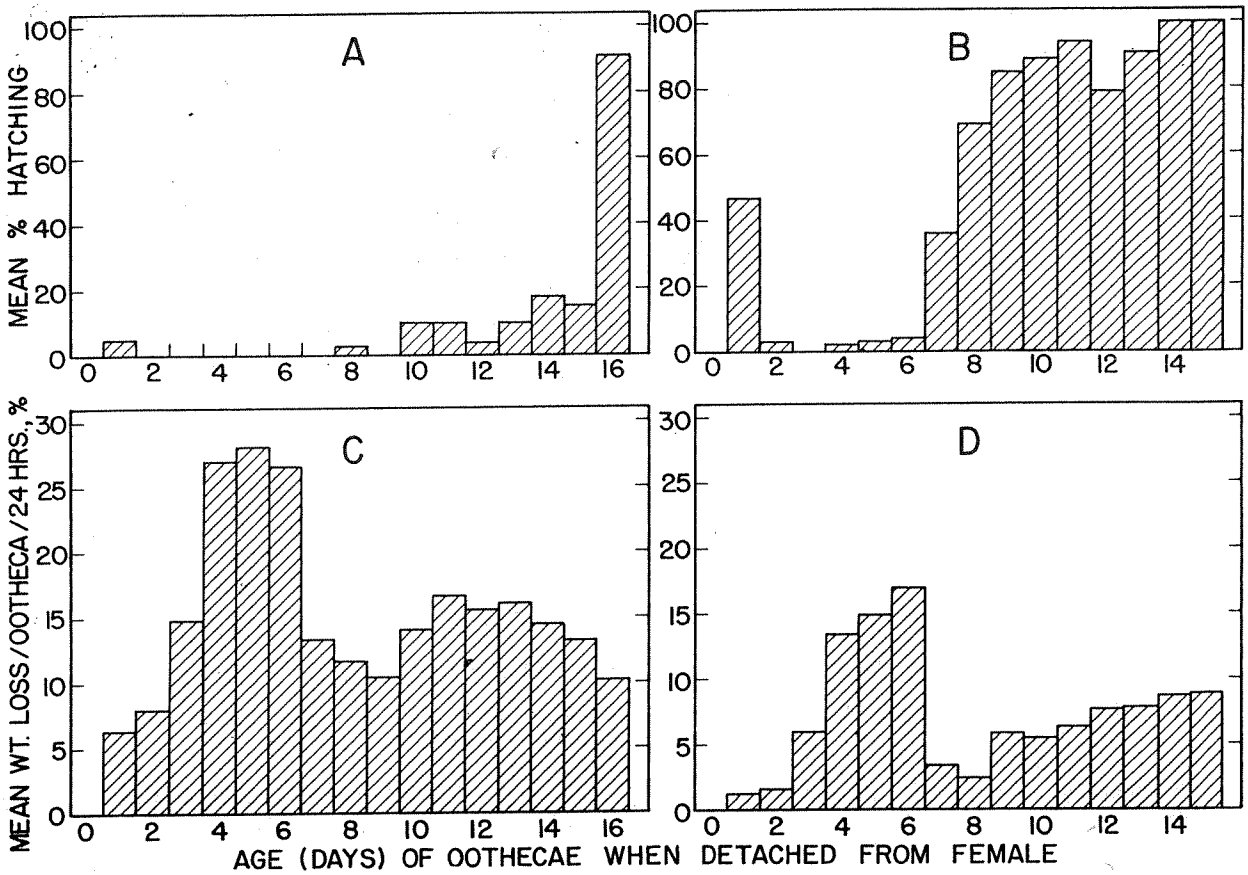

FIG. 1.—Effect of relative humidity on weight loss and hatching of eggs in manually detached oöthecae of *Blattella germanica*. A–B. Mean per cent hatching refers to the per cent of the oöthecae used from which eggs hatched. A = Oöthecae kept at 30% to 50% relative hunidity. B = Oöthecae kept at 90% relative humidity. (Data from table 1.) C–D. Mean weight loss, over a 24 hour period, of manually detached oöthecae. C = Oöthecae kept at 30% relative humidity; from 6 to 29 Oöthecae were used for each age group (total of 373 egg cases). D = Oöthecae kept at 90% relative humidity; from 6 to 24 oöthecae were used for each age group (total of 171 egg cases).

age hatch was high only when the egg cases were detached from the females one day prior to hatching. When oöthecae were kept at 90 per cent R. H., a high percentage hatch was obtained only if the oöthecae were 1-day or 7 to 14 days old when removed from the females; even at this high humidity there was little or no hatch from oöthecae that were 2 to 6 days old when detached. The fact that detached 1-day old eggs will develop and hatch at 90 per cent R. H. shows that the recently-formed eggs of the German cockroach contain enough water to complete development without the additional water which normally is picked up from the female during embryonic development; even at the high humidity water is not taken up by detached eggs, but both weight (Figs. 1D, 3) and water (as indicated by the marked drop in weight) are continuously lost.

Table 2.—**Effect of relative humidity on development and hatching of eggs from manually detached oöthecae of *Blattella germanica*.**

Age (Days) of Oöthecae When Detached	Mean No. Eggs per Oötheca	Per Cent Eggs per Oötheca Which:		Number of Oöthecae Used[b]
		Developed	Hatched[a]	
Control[c]	30.4	98.5	99.9	26
	Oöthecae kept at 30% to 50% R. H.			
1, 8, 10–14	36.1	95.2	69.6	23
15–16	34.4	94.1	88.4	40
	Oöthecae kept at 90% R. H.			
1–2, 4–8	32.8	94.8	83.6	52
9–15	32.0	98.3	93.3	135

[a] Based only on eggs which developed.
[b] Some eggs hatched from all oöthecae used.
[c] Oöthecae left on females until eggs hatched.

Eggs did not hatch from detached, 2- to 4-day old oöthecae kept at a low humidity; few of the eggs developed into recognizable embryos, and those that did were situated at the sclerotized posterior end of the oötheca. When 5-day old or older oöthecae were removed from the females, most of the eggs developed; those that did not develop were located at the soft anterior end of the oötheca. These results are attributed to the difference in permeability of the two ends of the oötheca; water is lost more rapidly from the anterior end of detached oöthecae (Roth & Willis 1955). Even though hatching did not oc-

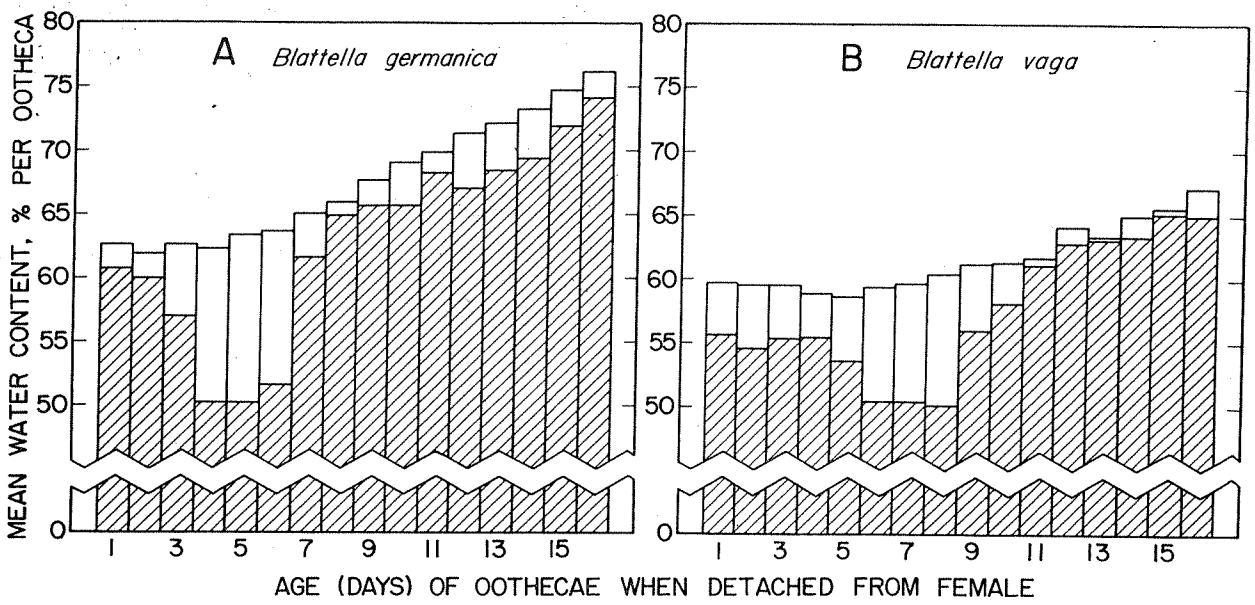

FIG. 2.—Water content of the oöthecae of *Blattella*. Open bars = Normal water content; from 20 to 30 oöthecae (total of 385) of each age group of *B. germanica*, and from 10 to 15 oöthecae (total of 220) of each age group of *B. vaga*. Hatched bars = Water content after 24 hours at about 30 per cent relative humidity; the same oöthecae of *B. germanica* used in figure 1C were used here and from 9 to 24 oöthecae (total of 227) of *B. vaga*.

cur, many eggs developed to mature embryos at 90 per cent R. H. regardless of the age at which the oötheca was detached. Of those oöthecae from which eggs hatched, the percentage of eggs which developed, per oötheca, was high, regardless of the age when detached or the humidity at which they were kept; however the percentage of eggs, per oötheca, which hatched was smaller at the low humidity, and at both high and low humidities the younger groups of detached eggs had a smaller per cent hatch than the older groups (Table 2).

It is evident that even at the high humidity there is a critical period, from the second to the sixth day, during which most of the eggs will not hatch if the oöthecae are detached from the females at that time, We kept 2- to 5-day old oöthecae on filter paper moistened with physiological saline solution, as recommended by Parker & Campbell (1940), and found that the eggs in 40 per cent to 54 per cent of the oöthecae hatched (Table 3); the percentage hatch would probably have been higher were it not for bacterial and fungal growth which was uncontrolled. Evidently the failure of eggs to hatch from oöthecae removed from females during this critical period is not due to injury to the eggs themselves, although the possibility that the oöthecae have been damaged in some way still remains; however, many 1-day old oöthecae were successfully removed without apparent injury.

Table 3.—Hatching of eggs of *Blattella germanica* from oöthecae kept in contact with physiological saline solution.

Age (Days) of Oöthecae When Detached	Number Used	Hatched		Age (Days) When Removed from Saline Solution and Kept at 90% R. H.
		Number	Per Cent	
2	20	8	40	13
3	7	3	43	14
4	24	13	54	15
5	21	10	48	16

The weight loss by oöthecae of different ages, over a 24-hour period, is shown in figure 1C and 1D. At about 30 per cent R. H. (Fig. 1C) the least amount of weight lost was from eggs in oöthecae that were 1-day old when removed from the females. The greatest loss occurred from oöthecae detached when they were 4 to 6 days old. At 90 per cent R. H. the amount of weight lost was less than that found in oöthecae kept at the lower humidity, but here again the loss varied with the age of the oötheca at the time of removal from the female; again 1-day old oöthecae lost the least weight and the 4- to 6-day old oöthecae lost the most. At 90 per cent R. H. the critical period during which little hatching occurred corresponds closely with the period during which the greatest weight was lost (cf., Figs. 1B and 1D). The greater weight loss at 30 per cent R. H. during the critical period was due to the greater water loss (Fig. 2A); this is undoubtedly true at 90 per cent R. H. although water determinations were not made of oöthecae kept at this humidity. The oöthecae of *Blattella vaga* Heb. lose weight and water in a similar manner; but the critical period during which the most weight and water was lost was 6 to 8 days in *B. vaga* (Fig. 2B) compared to 4 to 6 days in *B. germanica*.

It seems reasonable to conclude that the rate of water loss is determined by the age of the eggs at the time the oöthecae are removed from the females. The difference in

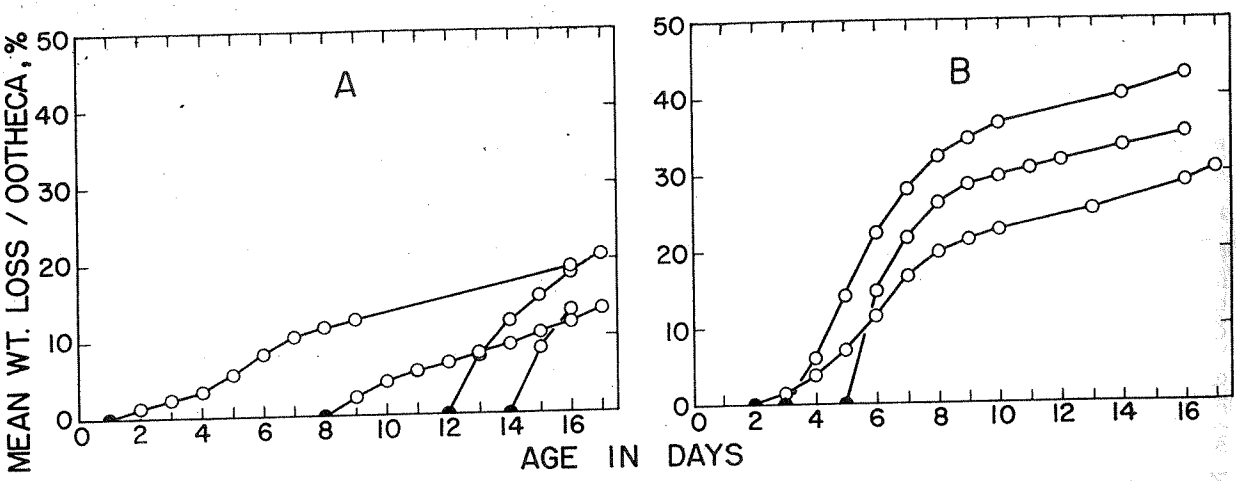

FIG. 3.—*Blatella germanica*. Effect of age, at time of removal from female, on rate of weight loss of manually detached oöthecae kept at 90 per cent relative humidity. Each curve starts (black semicircle) at the age the oöthecae were when removed from the females. From 6 to 15 oöthecae (total of 77) were used for each age group. In figure A some eggs hatched from each of the oöthecae whereas in B none of the eggs hatched.

time of occurrence of the critical period when the greatest amounts of water are lost can be correlated with developmental time of the eggs of *Blattella;* the eggs of *B. germanica* hatched in 17 days while those of *B. vaga* in 20 days. The weight changes in detached German cockroach oöthecae of different ages kept at 90 per cent R. H. were followed periodically from the time of their removal from the females until hatching (Fig. 3). It is interesting to note that although the rate of weight loss from 1-day old oöthecae increased somewhat when the eggs became 4 to 6 days old, the increase was not large enough to prevent the eggs from completing their development and hatching. If the high rate of water loss of 3- to 6-day old eggs were due to a particular stage of development of the egg (e.g., certain membrane changes such as are found in grasshopper eggs), one would expect a sudden increase in water loss when the eggs reached this so-called critical period in age. Perhaps the oötheca may be damaged in some way when removed at 2 to 6 days of age so that water is lost more rapidly.

The proportion of water in the oötheca and eggs of *B. germanica* is initially about 62 per cent by weight. In oöthecae carried by females this remains fairly constant for about 4 days and then gradually increases to about 75 per cent by the time of hatching, as dry weight is lost and water is absorbed from the female (Fig. 2A, open bars; Roth & Willis 1955). Whether or not the eggs in detached oöthecae will survive and hatch will depend on the rate of water loss and the amount of water present at the time of removal from the female. When kept at a high humidity some 1-day old eggs lose water so slowly (Fig. 3A) that they complete their development and hatch. Eggs in oöthecae detached when 2 to 6 days old lose water too rapidly (Fig. 3B) to hatch. Seven-day old or older eggs have begun to increase in water content; thus even though the rate of water loss in older eggs may be the same or greater than the 2- or 3-day old eggs (cf., Figs. 3A and 3B), hatching can take place in the older eggs because they have more water and have nearly completed their development. Unfortunately, practically nothing is known of the physical or chemical properties of cockroach egg membranes which control water loss; this lack of information makes it difficult to interpret the results of this investigation (cf. Salt 1952).

SUMMARY.—When kept at a high humidity, a large percentage of eggs in detached oöthecae of *Blattella germanica* hatch if the egg cases are 1 or 7 or more days old when removed from the females. When kept at a low humidity there is little or no hatch from eggs in detached oöthecae except in egg cases removed about 1 day prior to hatching. Hatching of eggs in detached oöthecae depends on the rate of water loss, the amount of water present in the eggs at the time the oötheca is removed from the female, and the amount of time remaining to complete development. The rate of water loss varies with the age of the oötheca at the time of detachment. The slowest rate of loss occurs in oöthecae which are 1-day old when detached, and the greatest rate of loss occurs in eggs which are 4 to 6 days old at the time of removal from the female.

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